AMENDMENTS TO SPECIFICATION:

Please replace the paragraph bridging pages 12-13 with the following paragraph.

Isolation of pleurocidin cDNA

A cDNA library constructed from winter flounder skin (Gong et al 1996) was screened using degenerate oligonucleotides (PleuroA, PleuroB; Table 1). The library was plated at 80,000 phage/plate and duplicate lifts to HyBond filters were made of each of eight plates. A mixture of radioactively end-labelled PleuroA and PleuroB probes was hybridised with the filters at 50° C using standard procedures, and the filters were washed in 1X SSC/0.1% SDS at 50° C for 45 min. Plaques that showed matching hybridization signals on both duplicate filters were picked and the library rescreened until 100% purity of the recombinant plaques was obtained. Two recombinants were completely sequenced using an ABI373 stretch automated sequencer and the AmpliTaqFS Dye Terminator Cycle Sequencing Ready Reaction kit (Perkin-Elmer, Foster City, CA, USA). Sequence data were analyzed using Sequencher (Gene Codes, Inc., Ann Arbor, MI, USA) and DNA Strider. The amino-terminal signal sequence was predicted using SignalP (~~http://~~ "www" followed by ".cbs.dtu.dk/services/SignalP"). The Helical Wheel routine of the GCG package (~~http://~~ "www" followed by ".gcg.com") was used to model the helical structure of the predicted antimicrobial peptide sequences.

Please replace the first full paragraph on page 16 with the following paragraph.

Identification of pleurocidin-like sequences in the winter flounder genome

A winter flounder genomic λ-GEM library was screened using a radioactively labeled probe for pleurocidin (WF2; Douglas et al., 2001). Four clones were picked and replated until 100% purity was achieved. The clones were mapped using *BamHI, SstI, XhoI* and *Eco RI* and two clones (λ1.1 and λ5.1) that differed in restriction pattern were selected for sequencing. Both clones were completely sequenced using an ABI373 stretch automated sequencer and the AmpliTaqFS Dye Terminator Cycle Sequencing Ready Reaction kit (Perkin Elmer, Foster City, CA, USA. Transcription factor binding sites were identified using

WWW Signal Scan (~~http://~~bimas.dcrt.nih.gov/molbio/signal/) with the TransFac and TFD databases and promoters were detected using the eukaryotic promoter prediction by neural network software available at the Baylor College of Medicine (~~http://~~searchlauncher.bcm.tmc.edu/seq-search/gene-search.html).

Please replace the paragraph bridging page 16-17 with the following paragraph.

Molecular Characterisation of Hepcidin cDNAs

Eight ESTs showing high similarity to human hepcidin were identified from the winter flounder EST database (Douglas, Gallant et al. 1999) and four from the Atlantic salmon database (Douglas, Tsoi et al. 2002). Using these sequences to screen dbEST, BLASTX analysis revealed two related sequences from Japanese flounder (C23298.1 and C23432.1), one sequence from rainbow trout (AF281354_1) and five identical sequences from medaka (AU178966, AU179222, AU179314, AU179768 and AU180044). Sequence data were analyzed using Sequencher (Gene Codes, Inc., Ann Arbor, MI, USA) and DNA Strider (Marck 1992). Alignments and similarity matrices were calculated using ClustalW (Thompson, Higgins et al. 1994) and graphically visualised using SeqVu (Garvan 1996). The on-line servers PSORT (~~http://~~ PSORT.nibb.ac.jp), Compute pI (~~http://~~expasy.hcuge.ch/cgi-bin/pi_tool), and Network Protein Sequence @nalysis (~~http://~~npsa-pbil.ibcp.fr/cgi-bin/secpred_consensus.pl) were used to predict N-terminal signal sequences, pI and secondary structure, respectively. The secondary structure prediction program utilized seven different algorithms (for details, see web site) and provided a consensus prediction based on these results.

Please replace the second full paragraph on page 21 with the following paragraph

In order to estimate the net charge, K and R were assumed to have the value of +1, H of +1/2, D and E of -1, and C-terminal amidation was counted as an additional +1.

The EMBOSS Pepwheel and Pepnet internet tools available through an NRC mirror site (~~http://~~bioinfo.pbi.nrc.ca:8090/EMBOSS/index.html) were used to

analyse the separation of hydrophilic and hydrophobic residues in helical wheel and helical net models.